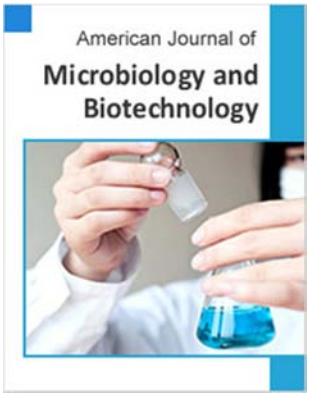
American Journal of Microbiology and Biotechnology
2017;4(1): 8-13
http://www.aascit.org/journal/ajmb
ISSN: 2375-3005

American Association for Science and Technology



Keywords
Proximate Analysis,
Soy Milk Yoghurt,
Cow Milk Yoghurt,
Composite Yoghurt,
Commercial Yoghurt,
Starter Culture

Received: March 17, 2017
Accepted: March 30, 2017
Published: June 7, 2017

Comparative Studies on the Nutritional and Physicochemical Properties of Yoghurts Produced from Soy and Cow Milk

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Citation
Ezeonu Chukwuma Stephen, Tatah Verwiyeh Silas, Iyua Iorlumun. Comparative Studies on the Nutritional and Physicochemical Properties of Yoghurts Produced from Soy and Cow Milk. *American Journal of Microbiology and Biotechnology.* Vol. 4, No. 1, 2017, pp. 8-13.

Abstract
In this study, biochemical composition and sensory evaluation of yoghurts from soy, cow and commercial milk was carried out. Proximate analysis carried out on samples of cow yoghurt (CY), soy yoghurt (SY), cow/soy yoghurt (CSY) and commercial yoghurts (CMY), showed significant difference ($P<0.05$) in crude protein of (SY) 6.27% and cow/soy yoghurt (CSY) 6.87%. However, there was no significant difference between CSY and CMY 7.09%. The crude fat in SY (4.44%) differs significantly with that of CMY (4.79%) but there was no significant difference between CSY (4.64%) and CMY (4.79%). The ash content of CMY (0.98%) was higher compare to CSY (0.65%). No significant difference was observed in the moisture content of the three samples, but SY had the highest value (8.29%) of carbohydrate content. The percentage lactic acid of the samples was found to be significantly different ($p<0.05$) between CMY (0.89%) and CSY (0.74%). SY and CY had no significant different in (%) lactic acid (0.63%) and (0.59%) respectively. The result shows that potassium, phosphorus, and calcium contents were found to be higher in CSY and SY when compared to CY. Vitamin A content in all sample were significantly different ($P<0.05$). Vitamin B_2 in SY (0.24mg/100mL) and CSY (0.25mg/100mL) had no significant different with CMY (0.21mg/100mL). Vitamin B_{12} in SY (0.35mg/100mL) had no significant difference ($p<0.05$) with CY (0.42mg/100mL), and CMY (0.36mg/100mL) samples. The result for sensory evaluation shows no significantly different ($p<0.05$) in colour of CY (7.10±1.06) and CSY (7.20±0.63. Taste and flavour in CY and CSY were significantly different ($p<0.05$). The result also shows that SY, CY, and CSY do not differ significantly in texture. The overall result showed that yoghurt from soy bean could compete favourably with the yoghurt from cow base-reference product. The % Nitrogen free extract and Energy values estimated showed that SY had the highest value compare to CMY.

1. Introduction

Historically, yoghurt was made by fermenting milk with indigenous microorganisms [1]. Nomadic people devised the production as an intuitive process to preserve the milk during travel [2]. Packaging was also an issue, they used animal skin to hold yoghurt and salted the product and thus made it more stable, texture wise and in preservation [2, 3]. During the fermentation, they started heating milk over an open fire in order to concentrate it slightly [4] to modify the properties of the casein, to eradicate any pathogenic microorganisms present in milk [5], to encourage the fermentation of milk to take place at a slightly higher temperature and also to ensure a gradual selection of lactic acid bacteria capable of tolerating high levels of lactic acid, and giving the product its distinctive flavour [6, 3]. Similar methods were used by the Turkish, Armenian, and Egyptians as well as other societies [6]. Each society found the best preservation methods for their needs [5], for instance, drying and heating for a few hours over low fires of a special type of wood that evolved a brand called smoked yoghurt, also yoghurt is preserved by keeping it salted or dried in olive oil or tallow. Another method that Turkish, Lebanese, Syrian, Iranian and Iraqi people used in milk preservation was mixing concentrated yoghurt with wheat to give a formulation called *kishk* [7]. After refrigeration became widespread, these traditional methods lost popularity except among certain communities in Middle East [8]. The production of yoghurt has increased due to its popularity as far as nutritional and therapeutic values are concerned [9]. Recently, yoghurt was tremendously popularized in Europe for its treatment of diarrhea, under the rule of Emperor Francis I of France. The methods of production over the years have changed bit by bit, for instance there is a trend of fruit yoghurts, but the fundamental steps remains the same [10]. The improvements in medical science research have also increased yoghurts nutritional effectiveness, resulting in its sustained popularity [11].

The presence of symbiosis of probiotics and prebiotics in yoghurt makes it highly functional food. Probiotics can be defined as "live microbial feed supplements that beneficially affect the host animal by improving its intestinal microbial balance" [12]. Prebiotics is a "non-digestible food ingredient that beneficially affects the host by selectively stimulating the growth and/or activity of one or a limited number of bacteria in the colon" [13]. The symbiosis "beneficially affects the host by improving the survival and the implantation of live microbial dietary supplements in the gastro-intestinal tract by selectively stimulating the growth and activating the metabolism of one or a limited number of health promoting bacteria" [14]. Under anaerobic environment, lactic acid bacteria (LAB) e.g. *Streptococcus thermophilus* and *Lactobacillus bulgaricus,* enzymes lactase degrade lactose to glucose and galactose from milk or milk products causing the formation of lactic acid in a product known as yoghurt. The sugar compound are then processed leading to the formation of lactic acid and acetaldehyde [15].

To produce yoghurt, milk is first heated, usually to about 85°C (185°F), to denature the milk proteins; eliminate pathogens and other unwanted or undesirable microorganisms [16]. After heating, the milk is allowed to cool to about 45°C. The bacterial culture is then mixed in the milk, and a temperature of about 45°C (113°F) is maintained for some hours (7hrs) to allow fermentation [5]. The nature and composition of yoghurt with its bacterial cultures determines the quality along with the nature of flavour and the way it appears [10]. The characteristic flavour of a yoghurt sample is due to the production of lactic acid, carbon dioxide, acetic acid, diacetyl, acetaldehyde and several other components from the milk fermentation process where the lactose is fermented by the lactic acid bacteria [17]. As a result, high priority is given to maintain good quality yoghurt, keeping in mind that even a small contamination can cause health disorders of consumers [18]. Until very recently, yoghurts have been made from various sources, including soy milk, grape juice, a combination of mango pulp–soy milk and buffalo milk, and merged with fruits such as natural fruit juice, pulp, dry fruits, and often to serve to increase the aesthetic value [19].

The characteristic taste of yoghurt is determined by its smooth, yet viscous with a subtle flavour that resembles a walnut [20]. The gel like texture is the primary characteristic and when added with thickening agent such as gelatin or other hydrocolloids, the yoghurt texture is shown to stabilize, leading to an effective resistance against syneresis while producing that smooth sensation for the mouth [10, 21, 22, 23]. The aim of this research was to comparatively determine the nutritional and physicochemical composition of yoghurts produce from soy, and cow milk, separately and in composite using commercial yoghurt as control.

2. Materials and Methods

2.1. Collection of Sample

Fresh cow milk was obtained from nomad in Wukari metropolis, Taraba State, Nigeria soybean seeds (variety TGX-923-2E) were purchased in Wukari market. The starter culture "Yogourmet" containing *Streptococcus thermophilus*, *Lactobacillus bulgaricus* and *Lactobacillus acidophilus* a product of Lyosan Inc. England, was obtained at a shopping complex along Kashim Ibrahim Road Makurdi, Benue State, Nigeria. Commercial yoghurt (Hollandian yoghurt) was purchased from a local store opposite Federal University Wukari and used as control sample.

2.2. Preparation of Soymilk

Soymilk was produced according to the method described by Lui, [24]. Selected (sorted) soybean seeds (0.5kg) was weighed and washed (surface-sterilized) with boiled water and then rinsed for 2minutes. The surface-sterilized seeds were soaked (steeped) in sterile distilled water in a sterilized container (5Litres) for about 5hrs at 25°C. The soybean seeds were then dehulled, thoroughly washed and further milled with distilled water for about 4 minutes using a HR2004 blender (Philips, Guangzhou, PR China). The resulting slurries were mixed with water and filtered through a milk filter to yield soymilk.

2.3. Yoghurt Production

Three milk samples were used for yoghurt preparation. Sample CY (cow milk yoghurt) was prepare from cow milk, samples SY (soymilk yoghurt) from soymilk and sample

CSY (cow/soymilk yoghurt) were blends of cow milk and soybean milk at the ratio of 50:50% respectively. Each sample was then pasteurized at 85°C for 20 minutes, cooled to 43°C, inoculated with 5% starter culture inoculums and finally incubated at 41°C for 48 hours to ferment. The fermented sample (yoghurt) were cooled and stored in a refrigerator for further physicochemical analysis and sensory evaluation.

2.4. Proximate Composition

Proximate analysis was carried out to determine the nutritional composition of the produced yoghurt. The Moisture, Ash, Protein, Crude fibre, fat and carbohydrate contents of the produced yoghurt was determined using the analytical method described by AOAC, [25]. Each parameter was analysed in triplicate for statistical analysis.

2.5. Mineral Analysis

Mineral analysis was carried out using dry digestion method. The method described by AOAC, [25] was adopted. Calcium, phosphorus, potassium were analysed from the triple acid digestion (wet digestion method). Each parameter was analyzed in triplicate for statistical analysis.

2.6. Vitamins Determination

Water and Fat Soluble Vitamins were determination by Isocratic High Performance Liquid Chromatography (HPLC). Each parameter was analyzed in triplicate for statistical analysis.

2.7. Determination of pH

The pH of yoghurt was determined after 24 hours and 21 days of production using a digital pH-meter (Jenway 3505, UK) calibrated with pH 4 and 7 buffers. Each pH value was determined in triplicate for statistical analysis.

2.8. Sensory Evaluation

Sensory properties of samples were evaluated by a trained panel consisting of 10 assessors (including students and staff in Food Science and Technology and Biochemistry Departments). The various yoghurt samples were served at 7 to 10°C in plastic cups and coded with three-digit numbers. Order of presentation of the samples was randomized. A test form (questionnaire) comprising five sensory attributes namely *viz;* colour, taste, flavour, texture and overall acceptability, was given to each assessor. A standard 9-point scale was used for evaluation of sensory characteristics of samples, in which 1 was equal to dislike extremely and 9 was equal to like extremely.

2.9. Gross Energy Determination

The energy in food is measured in calories (cal) and joules (J). The energy value of samples according to AOAC, (1990)[25] was determine by multiplying the % carbohydrate content by 4%, protein content by 4% and fat content by 9%. It was calculated thus;

Energy value = (% CP × 4) + (% CFT× 9) + (% NFE × 4)

While, % NFE (nitrogen free extract) = 100- (% CP + % CF + % CFT+ % ash + % moisture),

Where:

% CP = percentage crude protein

% CFT = percentage crude fat

% NFE = percentage nitrogen free extract

% CF = percentage crude fibre.

2.10. Statistical Analysis

The results obtained from proximate, minerals, vitamins and sensory analyses were subjected to Independent sample T-test using IBM SPSS (20 Version). Significance different between samples was tested at $p<0.05$.

3. Results and Discussion

The results of the proximate analysis of yoghurt samples are presented in table 1 below. From the results, the mean values of the crude protein were significantly different ($p<0.05$) between soymilk yoghurt (SY) (6.23%), cow/soymilk yoghurt (CSY) (6.87%) and cow milk yoghurt (CY) (4.50%) samples. The crude protein was found to be higher in commercial yoghurt (CMY) (7.09%) than in other samples, and it has no significant difference ($p<0.05$) with CSY (6.87%). The highest value in crude protein may be due to fortification of the yoghurt with amino acids [26]. The lowest mean value was observed in cow milk yoghurt (CY) (4.50%), which was significantly different with other yoghurt samples. There was no significant difference ($p<0.05$) in crude fat content between SY (4.44%) and CY (3.59%), and between CSY (4.64%) and CMY (7.09%). This signifies that CSY competed favourably with CMY [27]. The ash contents were found to be very low in all the samples as shown in table 1, with CMY having the highest value of 0.98%, which may be due to fortification of yoghurt with mineral elements. All samples had high moisture content above 81.0%. However, there was no significant difference ($p<0.05$) in moisture content between as shown in table 1. There was a significant difference ($p<0.05$) in carbohydrates contents between CSY (6.41%) and CMY (4.50%), as well as between SY (8.92%) and CY (4.68%). The value of carbohydrate in CY (4.68%) and CMY (4.50) were not significantly different at $p<0.05$. The high mean score value of carbohydrate in SY sample may be attributed where the plant was planted. The values obtained in this study for crude protein, fat, moisture content, ash and carbohydrates fall within the range obtained by Osundahunsi *et al.*,[26] and Orlowski *et al.*, [27] for soy yoghurts.

Table 1. *Proximate Composition of Yoghurt Samples (%).*

Parameters	SY	CY	CSY	CMY
Crude protein	6.23±0.01^{a}	4.50±0.01^{c}	6.87±0.01^{ab}	7.09±0.02^{ab}
Fat	4.44±0.01^{b}	3.59±0.01^{b}	4.64±0.00^{c}	4.79±0.01^{c}
Ash	0.89±0.01^{d}	0.65±0.00^{c}	0.97±0.01^{a}	0.98±0.00^{a}
Moisture	82.04±0.01^{a}	82.99±0.01^{a}	82.85±0.00^{a}	82.65±0.01^{a}
Carbohydrate	8.29±0.03^{d}	4.68±0.01^{c}	6.41±0.04^{a}	4.50±0.05^{c}

Values are means ± standard deviation from duplicate determinations
Means in the same row with the same superscript letter are not significantly different at ($p<0.05$)
KEY: SY=Soymilk yoghurt, CY=Cow milk yoghurt, CSY=Cow and soymilk yoghurt (i.e. Composite yoghurt), CMY=Commercial yoghurt (i.e. control).

The pH and titratable acidity of yoghurt samples are presented in table 2. The pH was determine after 24 hours of fermentation and after 21 days of storage in the refrigerator at the temperature range of 2-4°C. The pH of the yoghurt samples after 24 hours of fermentation (except sample CMY) showed that there was significant difference ($p<0.05$) in SY (4.53±0.01), CY (4.21±0.02), and CSY (4.34±0.02) samples respectively. Whereas, no significant difference ($p<0.05$) was observed between CY (4.21±0.02) and CSY (4.34±0.02), as well as between SY (4.53±0.01) and CMY (5.31±0.01) samples. This slight differences in pH could possibly be due to storage condition and low acidity. Sample CY had the lowest pH of 4.21, which show that it had high concentration of lactose which is an ideal substrate for yoghurt starter cultures. The pH values of the samples after 21 days of refrigeration show that, there were no significant difference ($p<0.05$) among SY (5.73±0.00), CY (5.96±0.00), and CSY (5.60±0.00 samples). This may be due to storage conditions (2-4°C) that inhibit the activity of the microorganisms to produce more lactic acid. The pH values of all the samples fall within the range of good quality yoghurt [28][29]. No significant difference was observed in the titratable acidity of SY (0.63±0.01) and CY (0.59±0.01). Whereas a significant difference was observed in sample SY (0.63±0.01), CSY (0.74±0.01), and CMY (0.89±0.01) respectively.

The mineral composition of the yoghurt samples presented in table 3 shows significant different ($p<0.05$) among the samples. Calcium (232.42mg/100g) and Phosphorus (203.34mg/100g) were high in CMY sample, probably because of the fortification of yoghurt with minerals. Potassium content was high in CSY (177.20mg/100g), followed by CMY (161.64mg/100g), SY (157.50mg/100g), and CY having the lowest value of 121.36mg/100g. The results of mineral content also shows that sample CSY competed favourably with sample CMY in terms of Calcium and Phosphorus. Whereas, Potassium was found to be higher in CSY (177.20mg/100g) compared to sample CMY with the Calcium content of 161.54mg/100g.

The composition of vitamins in the yoghurt samples are shown in table 4. The highest vitamin A content was observed in sample CY with the mean value of 61.54IU/ML, and the lowest mean value was observed in sample SY (51.77IU/mL). There was a significant different ($p<0.05$) in vitamin A content between SY (51.77IU/mL), CY (61.54IU/mL), CSY (54.83IU/mL), and CMY (57.71IUmL) respectively. Sample CY had the highest value of vitamin B2 (0.29mg/100mL), the lowest value was observed in CMY (0.21mg/100mL). From the results, a significant different existed between SY (0.24mg/100mL) and CMY (0.21mg/100mL), CY (0.29mg/100mL) and CSY (0.25mg/100mL) respectively. Vitamin B12 was found to be higher in sample CY (0.42mg/100mL), and lower in sample CSY (0.33mg/100mL). There was no significant different ($p<0.05$) in sample SY (0.35mg/100mL), CY (0.42mg/100mL), and sample CMY (0.36mg/100mL). The value of sample CSY differed from other samples.

The results for sensory evaluation are presented in table 5. The result shows that, there is no significant different ($p<0.05$) in the colour of CY (7.10±1.11) and CSY (7.20±0.63), but there was a significant different ($p<0.05$) in colour of sample CSY and CMY (8.00±1.05), SY (6.30±1.06) and CY (7.10±1.11), CY (7.10±1.11) and CMY, as well as between SY and CMY respectively. The highest mean score for colour was observed in sample CMY, followed by CSY, CY, and SY as shown in table 5. This result shows that SY compete favourably with CMY and surpass CY samples. Hence sample CSY can be presented as substitute for CY and CMY in term of colour. The CMY has the highest mean score for taste (8.40±0.52), which is significantly different ($p<0.05$) from other samples. This is probably due to the addition of taste enhancers in CMY sample. There was no significant different ($p<0.5$) between the taste of sample SY (6.50±1.51), and CY (6.80±31). Sample CSY (6.10±1.97) had the lowest mean score.

The CMY sample had the highest mean score (8.30±0.82) for flavour, which is significantly different ($p<0.05$) from other samples as shown in table 5, but there was no significant difference ($p<0.05$) between SY (6.50±1.18) and CY (6.70±1.34). The lowest mean score for flavour was observed in CSY (6.10±1.97). This might be due to the beany flavour of soybean blended with cow milk [30]. The mean score for texture show that, there was no significant different in CY (6.90±1.45), SY (7.00±0.82), and CSY (7.00±1.56) samples. The highest mean score for texture was observed in CMY (8.10±0.88), which is significantly different ($p<0.05$) from other samples. The lowest mean score was observed in CY (6.90±1.54). This was probably because of the high moisture content (82.99%) of CY, which is related to syneresis [31]. The mean score for overall acceptability of the samples shows that, sample CMY has the highest mean score of 8.40±0.61, which is not significantly different ($p<0.05$) from CY (7.50±1.18) sample. The lowest mean score was observed in sample CSY (7.30±1.42). There was a significant different ($p<0.05$) in the overall acceptability of

SY (7.40±0.84) and CMY (8.40±0.61). SY (7.40±0.84) and CSY (7.30±1.42) were the least accepted samples in sensory analysis. This was probably because of the astringency and beany flavour associated with soybean [23].

The results obtained showed that there was significant difference ($p<0.05$) across the samples in terms of % nitrogen free extract with sample SY having the highest value of 6.40±0.01, and sample CMY having the lowest value of 4.49 ±0.01. The energy values (Kcal.) showed that there was a significance difference ($p<0.05$) among all the samples with sample SY having the highest energy value of 90.48Kcal/100g, and sample CY having the lowest energy value of 83.23Kcal./100g.

Table 2. *pH and Titra table Acidity of Yoghurt Samples.*

SAMPLES	pH (after 24 hours)	pH (after 21 days)	Titratable (% Lactic acid)
SY	$4.53±0.01^{b}$	$5.73±0.00^{b}$	$0.63±0.01^{c}$
CY	$4.21±0.02^{a}$	$5.96±0.00^{b}$	$0.59±0.01^{c}$
CSY	$4.34±0.02^{a}$	$5.60±0.00^{b}$	$0.74±0.01^{d}$
CMY	$5.31±0.01^{b}$	$5.43±0.01^{a}$	$0.89±0.01^{ad}$

Means with the same superscript letter within the same column are not significantly different at ($p<0.05$).

Table 3. *Mineral Composition of Yoghurt Samples (mg/100g).*

SAMPLES	Potassium (K)	Phosphorous (P)	Calcium (Ca)
SY	$157.50±0.01^{a}$	$179.32±0.01^{c}$	$193.58±0.01^{a}$
CY	$121.36±0.01^{b}$	$153.38±0.01^{ab}$	$177.32±0.01^{c}$
CSY	$177.20±0.03^{c}$	$187.95±0.12^{b}$	$208.49±0.12^{b}$
CMY	$161.54±0.01^{d}$	$203.34±0.12^{ac}$	$232.42±0.01^{d}$

Means with the same superscript letter within the same column are not significantly different at ($p<0.05$).

Table 4. *Vitamins Composition of Yoghurt Samples (mg/100g).*

SAMPLES	Vitamin A (IU/mL)	Vitamin B2 (mg/100mL)	Vitamin B12 (mg/100mL)
SY	$51.77±0.01^{b}$	$0.24±0.01^{a}$	$0.35±0.01^{b}$
CY	$61.54±0.01^{a}$	$0.29±0.01^{b}$	$0.42±0.01^{b}$
CSY	$54.83±0.01^{c}$	$0.25±0.01^{b}$	$0.33±0.01^{c}$
CMY	$57.71±0.02^{d}$	$0.21±0.01^{a}$	$0.36±0.01^{b}$

Means with the same superscript letter within the same column are not significantly different at ($p<0.05$).

Table 5. *The Result of Sensory Evaluation Attributes.*

SAMPLES	Colour	Taste	Flavour	Texture	Overall acceptability
SY	$6.30±1.06^{a}$	$6.50±1.51^{a}$	$6.50±1.18^{d}$	$7.00±0.82^{b}$	$7.40±0.84^{a}$
CY	$7.10±1.11^{b}$	$6.80±1.31^{a}$	$6.70±1.34^{d}$	$6.90±1.45^{b}$	$7.50±1.18^{c}$
CSY	$7.20±0.63^{b}$	$6.10±1.97^{ab}$	$6.10±1.97^{ab}$	$7.00±1.56^{b}$	$7.30±1.42^{a}$
CMY	$8.00±1.05^{c}$	$8.40±0.52^{c}$	$8.30±0.82^{c}$	$8.10±0.88^{c}$	$8.40±0.61^{c}$

Means with the same superscript letter within the same column are not significantly different at ($p<0.05$).

Table 6. *The results for the gross energy determination presented in their mean ± standard deviation.*

Samples	% Nitrogen Free Extract	Energy Value (Kcal/100g)
SY	$6.40±0.01^{a}$	$90.48±0.02^{b}$
CY	$8.23±0.01^{b}$	$83.23±0.01^{d}$
CSY	$4.65±0.00^{c}$	$87.84±0.00^{c}$
CMY	$4.49±0.01^{d}$	$89.43±0.01^{a}$

Means with the same superscript letter within the same column are not significantly different at ($p<0.05$).

4. Conclusion

This research showed that yoghurt samples produced from soymilk and in combination of soymilk with cow milk will compete favourably with yoghurt produced from cow milk and the fortified commercial yoghurt. Nutritionally, the yoghurt met the dietary requirement of pure yoghurt without significant difference. However, flatulence factor and objectionable flavour in soybean products must be reduced or eliminated to enhance acceptability. Following the result of analysis carried out in this work, it is recommended that: There is need for more research on how to mask the beany flavour of soymilk to produced highly acceptable soy yoghurt. There is need to improve the values of soy yoghurt and soy/cow milk yoghurt in terms of colour, tastes, and flavour, by choosing appropriate flavour or other additives with low side effects which could surely enhance soy yoghurt acceptability.

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